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No. 3

- I. THE BLOOD OF INSECTS, WITH SPECIAL REFERENCE TO THAT OF THE COMMON HOUSEHOLD GERMAN OR CROTON COCKROACH, *BLATTELLA GERMANICA* LINN.*
- II. THE APPEARANCE OF LIPOMICRONS (MICRO FAT PARTICLES) IN THE BLOOD OF *BLATTELLA GERMANICA* LINN.
- III. THE TRACHEAL SYSTEM OF THE GERMAN COCKROACH, *BLATTELLA GERMANICA* LINN.

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To the Department of Entomology of Cornell University the investigator is especially indebted for the assignment of a desk at which to work and for the courtesies shown during the investigations.

The cockroaches which were used in these investigations were caught in the natural habitat in a trap wherein they could not be injured, Fig. 1. Specimens which long were kept in captivity were confined in special cages, such as is shown in Fig. 4. The moisture of the air in such a cage was kept so high that there was a fine condensation upon the walls of the cage bottles at all times, and the temperatures were as stated in various parts of the investigation records.

ANALYSIS OF COCKROACH BODY.

Ordinarily, normally the body of a cockroach contains a high percentage of water.

* A resume of a thesis which was presented to the Faculty of the Graduate School of Cornell University in partial fulfillment of the requirements for the degree of Doctor of Philosophy. June, 1924.

The average percentage contents of different substances in the body of the cockroach are as set forth in the following table. There were thirty-one of the younger nymphs and they were about three days old when the investigations were made. Two older nymphs which were about ready to make the final molt, two adult males and two adult females were used. A comparative table of the averages obtained is as follows:

TABLE No. 3.

	Average water per cent.	Average organic matter per cent.	Average ash per cent.
Adult males	68.84	30.19	.0942
Adult females	66.78	31.76	1.135
Older nymphs	66.85	31.95	1.186
Younger nymphs	79.21	17.90	2.88
Grand averages	70.24	27.95	1.535

The foregoing investigations were made merely to see how the results would check with those obtained from similar experiments of other investigators. Bodine, 1893-1921, working with certain species of grasshoppers obtained similar results with young and matured material as did Babcock, 1912, with clothes and bee moths.

FACTORS INFLUENCING SELECTION OF HABITATS BY COCKROACHES.

Later on in the thesis it will be seen that the ease with which cockroaches bleed is influenced by the relative humidity of the habitat from which they come, etc.

From some thirty observations made upon temperature and relative humidity in five separate natural habitats, it was revealed that the average temperature was 20.6 degrees C. or 69.08 degrees F. and the relative humidity was 71.9 per cent. By experimentation with a T-tube it was learned that if one end was supercooled while the other was superheated, the temperature of the air in the tube exactly midway between the superheated and the supercooled ends was half the difference between that of the two ends. It was then assumed that after the air in the middle of the tube was as stated, then from one end of the tube to the other, the temperature of the air inside gradually approaches that of either extreme. Then apparatus was fitted up to test out distribution of cockroaches in a tube the relative humidity and temperature of the air of which was known. The relative humidity was obtained

and maintained by bubbling air through distilled water for 100 per cent., by passing it through a saturated sodium chloride solution for a relative humidity of about 70 per cent. and by forcing it through concentrated sulfuric acid for less than 1 per cent. relative humidity. The jet of air was finely broken up as it passed through the respective solutions, so that the air automatically lost moisture to or took it from each respective solution, each giving a different relative humidity percentage to the air which was bubbled through it. The above mentioned materials are such as were used by Dr. Headlee, communicated to Dr. Wm. A. Riley in a letter in 1919. A glass tube which was about two feet long and one and one-sixteenth inches in diameter was selected. It was divided into eleven equal zones throughout its length, and the temperature per zone from the cooled to the heated extreme readily was calculated. A diagram of the apparatus is shown in Fig. 5. It was interesting to note that when the relative humidity was about 70 per cent. the cockroaches grouped themselves in zones the temperatures of which averaged 22.5 degrees C. or 72.5 degrees F. Some of the individuals left the zones of more favorable temperature and relative humidity, but it was with considerable caution, and even then they soon returned.

This experiment further revealed that, within reasonable limits, at lower temperatures cockroaches of this species seek seclusion in places the relative humidity of which is higher as compared to places in which the temperature is higher, for in the latter they sought seclusion in quarters the relative humidity of which was comparatively low. In other words, within reasonable limits, the lower the relative humidity, usually the warmer the seclusion quarters sought by *Blatella germanica* Linn., or the higher the relative humidity, usually the cooler the places of seclusion sought by them. When the relative humidity was too low, the cockroaches lost water to the surrounding air, as was shown by condensation of moisture upon the walls of the tube where the individuals were massed. In this respect my results agree with those of Bodine, 1893-1921, as he worked with grasshoppers.

To preclude the influences of light upon the behavior of the cockroaches, the experiments were made in a photographic dark-room, the lights of which were turned off excepting at times during the taking of readings, observations, the arrangement of apparatus and of the material.

It is believed that the foregoing discoveries offer an explanation for cockroaches seeking comparatively warm, moist seclu-

sion quarters in the natural habitat. The wax glands produce a waxy secretion, Dusham, 1918, which presumably keeps the body of proper moisture under ordinary circumstances. Cockroaches which were exposed until such moisture was lost bled with as much difficulty as did starved individuals.

In fifty-seven experiments in which the relative humidity was about 70 per cent. it was found that due to cold cockroaches became inactive at from 0 to 10 degrees C., the average being 5.02 degrees, at an average thermal range of 6.9 degrees C. over an average length of time of four minutes and thirty-six seconds.

In twenty-seven experiments in which the relative humidity was about 70 per cent. it was found that due to excessive heat cockroaches became inactive at from 40 to 54 degrees C., the average being 48.1 degrees C. at an average thermal range (twenty-two cases) of 14 degrees C. over an average length of time of four minutes and eighteen seconds.

Cockroaches were more passive to colder temperature extremes when the relative humidity of about 70 per cent was maintained at a constant. Those which became inactive as a result of exposure to cold became active more readily when returned to the natural habitat than did those which became inactive as a result of exposure to heat. Cockroaches could better withstand exposure to extremes of cold over extensive time intervals than they could to extremes of heat. Some which became inactive as a result of exposure to heat extremes remained so for several days, but finally again became active. Others were killed as a result of such exposure, and upon them a white fungus developed after several days.

INSECT BLOOD

Landois, 1864, experimented with fourteen different species of insects, and as a result of his investigations concluded that insect blood characteristically is of alkaline reaction, but Poulton, 1885, learned that the blood of phytophagous larvae and their resultant pupae (Lepidoptera) reacts acid to neutral litmus; Muttkowski, 1923, says that insect blood reacts alkaline or neutral to moist litmus, but that the reaction changes to acidic just before death. The investigator tested the blood of forty-two different species of insects which he recently had captured in the field. An antenna or a leg was clipped and the forthcoming blood droplet was tested with neutral litmus. As a result of his investigations he has concluded as follows:

Of all Orthoptera, Homoptera, Heteroptera, boring larval Lepidoptera, two phytophagous larval Lepidoptera and of two Coleoptera, one of the family Cerambycidae, the other of the family Carabidae, the blood reacted alkaline to neutral litmus.

Of all phytophagous Coleoptera, pollen eating Coleoptera, carnivorous Coleoptera and phytophagous larval Lepidoptera excepting two species, the blood reacted acid to neutral litmus.

Of all Diptera which were examined the blood reacted alkaline to neutral litmus. The blood of larval *Carpocapsa pomonella* reacted alkaline to neutral litmus, but the sap of the apple pulp upon which it was feeding gave a decided acid reaction.

The blood of certain phytophagous larval Lepidoptera reacted alkaline, while that of certain others reacted acid. The blood of a pollen eating beetle, *Cyllene robiniae*, a cerambycid, reacted alkaline; while that of others, *Chauliognathus*, a lampyrid, and of *Trirhabda*, a chrysomelid, reacted acid.

The blood color of representatives of different families of the same order of insects may differ, although the individuals feed upon the same host species. The blood of certain carnivorous Coleoptera (Coccinellidae), reacts acid and is of reddish, orange or yellowish color; while that of others (Carabidae) reacts alkaline and is of turbid white color.

The blood of adult phytophagous Coleoptera which came under observation is of yellow color, while that of phytophagous larval Lepidoptera feeding upon the same host species as those of the Coleoptera usually is of greenish color.

The foregoing results indicate that blood color modifications are of little taxonomic value, a fact which was pointed out by Milne-Edwards, 1835-36; and later by Poulton, 1885.

The results of these experiments show that influences other than the color of host sap act in the coloring of the blood, for insects of different species feeding upon hosts of the same species have blood which may present color differences and which may be of different color than is that of the host sap or tissues upon which the insects feed; that insects of different species feeding upon hosts of the same species may have blood which reacts differently and that the reaction of the blood may be different from that of the sap or the tissues upon which the insects feed. The results of the foregoing researches are in accord with those of Landois, 1864, and of Poulton, 1885.

The investigator agrees with certain others, that at least certain immature insects bear more blood in proportion to body weight

than do adults, for from adult individuals of *Pontia rapae* little blood was obtainable, whereas larval individuals of the same species yielded blood in profuse abundance. Others who have reached similar conclusions are Landois, 1864, and Miall and Denny, 1886.

According to the foregoing it is believed that the acidity, alkalinity and the color of insect blood must be species specificities and are influenced by more than just the food which the insect takes. Living insects of the same species in the same life cycle phases normally have blood of the same chemical (acid or alkaline) reaction, and usually of the same color.

The following excerpt from table No. 6 may be of interest in substantiating what already has been stated, as well as what is to follow:

TABLE No. 6.

HOST	INSECT	ORDER	FAMILY	REAC.	REMARKS
<i>Robinia</i>					
<i>pseudo-acacia</i>	<i>Cyllene robiniae</i>	Coleoptera	Cerambycidae	Alk.	Watery clear
.....	<i>Ecdytolopha</i>				
.....	<i>insiticiana</i>	Lepidoptera	Tortricidae	Alk.	Whitish
.....	<i>Epargyraeus</i>				
.....	<i>tityrus</i>		Hesperiidae	Alk.	Clear Green
<i>Asclepias syriaca</i> ..	<i>Euchaëitis egle</i>	Lepidoptera	Lymantriidae	Acd.*	Amb. Yellow
.....	<i>Tetraopes tetra-</i>				
.....	<i>ophthalmus</i>	Coleoptera	Cerambycidae	Acd.	Amb. Yellow
.....	<i>Anosia plexippus</i> ...	Lepidoptera	Nymphalidae	Alk.*	Clear Green
.....	<i>Doryphora clivi-</i>				
.....	<i>collis</i>	Coleoptera	Chrysomelidae	Acd.	Amb. Yellow
<i>Solidago cana-</i>					
<i>densis pollen</i>	<i>Cyllene robiniae</i>	Coleoptera	Cerambycidae	Alk.	Watery clear
.....	<i>Trirhabda</i>		Chrysomelidae	Acd.	Yellow
.....	<i>Chauliognathus</i>		Lampyridae	Acd.	Amb. Yellow

In the foregoing table, Reac. means the reaction to neutral litmus; Alk. signifies an alkaline and Acd. an acid reaction to neutral litmus; Amb. means amber, and the * means that the action was faintly as indicated.

Of the insects which fed upon black locust at some time during their development, it is interesting to note the wide color differences in the blood, and that all reacted alkaline to neutral litmus. See under *Robinia pseudo-acacia*.

Of those which feed upon milkweed, *Asclepias syriaca*, there is a difference in blood color and in reaction to neutral litmus. Of the Coleoptera, one of the family Chrysomelidae, another of the family Cerambycidae, the blood colors and the reactions to neutral litmus agree.

Of those taken while feeding upon the pollen of goldenrod, *Solidago canadensis*, all Coleoptera, there were differences in blood color and in reactions to neutral litmus.

Long ago the early microscopists, Malpighi and Leeuwenhoeck saw that the blood consists of formed bodies and of fluid plasma. Ordinarily the constituents of the blood plasma and those of the corpuscles are difficult to separate by hard and fixed lines, for it seems that some of the products of each are more or less interchangeable, as has been revealed by the researches of Newport, 1845, Wooldridge, 1884-5, Haycroft and Carlier, 1888, Cuénot, 1896, Nicholls, 1906, Lübben, 1907, Crawley, 1909 and subsequent workers. There is little doubt that the blood corpuscles draw their nourishment from the blood plasma. Edwards, 1835-6, Cuénot, 1896, and Lübben, 1907, believed that the vitalizing properties of blood are due to the corpuscles. Newport, 1845, and Cuénot, 1896, regarded them as floating, secreting glands. Newport, 1845, wrote that the blood continuously is changing due to the agency of the corpuscles and that the corpuscles are nourished by the blood plasma. Stokes and Wegefarth, 1897, believed that the granules of blood dust are shed from the leucocytes, Nicholls, 1906, regarded them as of diverse origin, and Crawley, 1909, believed that they come from erythrocytes.

Ordinarily the blood corpuscles of a cockroach are of opaque white color, this due to the white granules which they contain. In several instances it was noticed that the corpuscles were comparatively clear or hyaline and that at the same time the plasma was more heavily charged with white granules which appeared very much like those ordinarily noticed in the corpuscles. Although the writer did not see such granules leave the corpuscles, he is inclined to believe that those in the plasma come from the corpuscles.

In the following experiments each individual cockroach was kept confined in a small straight vial which was provided with a perforated screened cork, 1, 2 and 6 of Fig. 6. The perforation was provided with a loosely fitting bit of saturated sponge when it was desired to maintain a high relative humidity in the vial, see s of 6 of Fig. 6. Furthermore a small glass plate, 4 of Fig. 6,

with width equal to the inside diameter of the vial was placed into it, and beneath it was flooded with water, 5 of Fig. 6. If desired, food was placed into such an individual cage, otherwise a cockroach confined in it could eat nothing, but could get any desired amount of water. For experimenting in the natural habitat, no extra water was supplied (there was no sponge in the perforated cork and no water beneath the glass plate). If provided with only dry food in a dry habitat, in several days or even in shorter periods, depending upon the relative humidity and the thermal status of the air of that habitat, it became difficult to obtain blood from individuals thus kept. To substantiate the foregoing statement, Newport, 1845, Miall and Denny, 1886, Bruntz, 1908, and Muttkowski, 1923, state that blood quantity varies according to the nutrition status of the individual. My investigations show that in an otherwise favorable habitat with water always at their disposal, but absolutely starved with regard to other foods some cockroaches bled almost as freely and as copiously after eighteen days as they did at the outset of the experiment. Some of the individuals were bled several times during the above stated interval. All had starved to death at the end of twenty-one days with nothing but water at their disposal at a relative humidity of 100 per cent. and at an average temperature of 22 degrees C. or 81 degrees F. Usually males died first, followed by younger nymphs, older nymphs and adult females in the given order.

Ordinarily to the unaided eye, the fresh whole blood of the German or Croton cockroach may have a clearness like that of water, or but a slight to a comparatively heavy turbidity or cloudiness, depending upon how recently an individual has taken food. When placed against a clear white background, it can be seen that the basic color of the blood of this species is of rather a faint or delicate yellow. When set aside to clot and crystallize, the dried droplet takes on a deeper yellowish tinge, a highly perceptible glossy appearance, and melanosis has not yet developed in uncovered droplets which have been dried for almost a year. Landois, 1864, saw that fresh and dried blood of the same individual have the same basic color. Poulton, 1885, found that melanosis in insect blood is due to oxidation.

The whole blood of cockroaches consists of plasma and corpuscles. Milne-Edwards, 1835-6, reported that blood consists of water, fibrin, albumin, fatty phosphates, pigments, alkaline salts, cholesterin, urea, etc. To this Landois, 1864, added globulin and

metallic iron; Poulton, 1885, found pigments in the blood. He believed that they are not of respiratory nature and stated that when once they gain entrance they need not be renewed, he also found a reducing agent present in insect blood; MacMun, 1886, found myohaematin, the coloring agent of muscular tissue in blood and believed that it has to do with respiration; Griffiths, 1891, among certain of the more common constituents reported iron oxide, calcium, magnesium, soda, phosphoric acid, sulphuric acid, chlorine, oxygen, carbon dioxide and nitrogen. He says that the blood of Lepidoptera is rich in proteids which may separate out and retain much of the oxygen; Cuénot, 1896, believed that albuminoids of blood have respiratory and nutrient functions; Deegener, 1913, lists lutein; Muttkowski, 1923, besides more common substances reported gelatin, fibrinogen, fats, sugars, waste materials, enzymes, copper, potassium, magnesium, uranidine, iodine, lead and arsenic, the two latter regarded as accumulative. Griffiths, 1891, and Muttkowski, 1921, believe that copper in the blood of invertebrates acts as does iron in the blood of higher animals, the copper forming the nucleus of a respiratory protein, haemocyanin, as does iron in haemoglobin. Rollet, 1861, Lankester, 1867, MacMun, 1886, Cuénot, 1891, Griffiths, 1891, Berlese, 1909, and Vaney in larval *Gastrophilus*, Muttkowski, 1920-1, and Hungerford, 1922, report the presence of respiratory proteins in insects. I have seen crystals in especially prepared cockroach blood droplets which suggested those of hematin.

In making a spectroscopic examination of dried whole blood of the cockroach, Dr. Papish found that it contains iron, magnesium and sodium in relatively large quantities (as main constituents), calcium as a trace, aluminum as a small trace; potassium, titanium, copper and zinc in very faint traces (see Fig. 12).

In making a biochemical examination of fresh whole blood of the cockroach Dr. Bodansky found proteins in profuse abundance, especially fibrin; serum globulin and serum albumin are present to a marked degree; sugar from .10 to .12 per cent.; chlorides profuse; calcium oxalate a small amount; urea and uric acid doubtless present, but the available amount of blood too scanty to work out the percentages. Later the writer demonstrated the presence of a catalytic agent in the blood of the cockroach.

Landois, 1864, working with fourteen species of insects decided that each species of insect has its peculiarly characteristic

blood crystals. He regards them as being of organic nature and of both plasmal and corpuscular origin. Several kinds of crystals occur in an ordinarily desiccated cockroach blood coagulum. They are of various sizes and shapes, commonly of from six to fifty micra long, although some have been seen which were as many as one hundred sixty-four micra in length and almost as wide. It seems that among them are many sodium urate, uric acid, lysine and calcium carbonate crystals (Fig. 9). Like Landois, 1864, the investigator believes that blood crystals originate in blood corpuscles and from the plasma. In the dried droplet they are far more numerous than are the corpuscles, and of rather uniform distribution throughout the dried mass. If they originate only from the blood corpuscles, it would seem that they would be of more restricted or localized distribution in the droplet, that is, seemingly they would occur more or less grouped about each individual corpuscle. The exact nature of blood crystals has not been learned. Landois, 1864, believed that they are of organic nature, while Graber regarded them as being of inorganic nature. Glaser, 1916-7, agrees with Landois, for he says that they are of protein nature. In the small amount of blood which could be obtained from this species of cockroach, it was impossible to separate the plasma from the corpuscles before coagulation had set in. Blood crystals do not appear until after coagulation and drying to hardness, and I like Landois have seen them in especially prepared, stained ruptured corpuscles. They are regarded as being at least partly of organic origin. The following is the caption to Fig. 9:

A, measured from 17.1 to 22.8 microns, average 19.09 microns, of very slight yellowish green color, rather common in the microscopic field; probably sodium urate crystals.

B, measured from 20.8 to 37 microns, average 28.96 microns; of very yellow color, rather common; probably uric acid crystals.

C, measured from 47 to 164.5 microns, average 96.98 microns, of translucent yellowish tinge, rather common; may be lysine.

D, measured from 13.2 to 19.2 microns, average 16.48 microns; rather delicate appearing, often difficult to discern, rather common; probably calcium carbonate.

E, measured from 8.5 to 14.25 microns, average 11.10 microns; rather common, delicate appearing, suggestive of calcium carbonate.

F, measured from 6 to 16 microns, average 10 microns; very delicate, most common of all crystals; often difficult to see unless

focus and illumination are exactly right. Thus far I have been unable to identify these crystals.

In their normal living state the tissues of the cockroach are moist, this moisture being due to a serum which doubtless is a blood plasma derivative. Of the cockroach it gives a faintly alkaline reaction with neutral litmus. Due to this tissue serum it is difficult to learn the exact blood volume of an insect. Usually the bodies (muscles and other tissues) of younger, normally growing or developing individuals contain more water in proportion to their weight than do those of adult individuals of the same species. The results of Bodine's experiments with grasshoppers and those of Babcock with clothes and bee-moth larvae and adults agree with mine. The blood mostly consists of water. Just how much of the body moisture is serum in the tissues or what amount is plasma (in the blood stream) never has been satisfactorily settled for insects.

Edwards, 1835-6, saw that the proportion of plasma to formed bodies varies greatly in individuals of the same species; Newport, 1845, Miall and Denny, 1886, Bruntz, 1908, and Muttkowski, 1923, state that blood quantity varies according to the nutrition status of the individual. Cockroaches which were completely starved for several days and those which had lost much metabolic water were difficult to bleed, due to the scarcity of blood plasma. Of a well fed female from the natural habitat there was much more blood which was easily obtainable and it contained about twice as many corpuscles per unit volume as did that of some which were completely starved for twenty-four hours in the same habitat where the temperature was 22.5 degrees C. or 72.5 degrees F. and the relative humidity was 91.2 per cent. According to the foregoing it seems that the number of corpuscles in the blood lessens with starvation prolongation.

Bowerbank, 1834, in Ephemeroidea, Newport, 1845, in caterpillars (none in adults), Moseley, 1871, in *Blatta orientalis* (male figured), Deegener, 1913, Tillyard, 1917, and Muttkowski, 1924, record having seen oat- or spindle-shaped corpuscles in the blood of various insects with which they worked. If a form is sufficiently transparent that the blood can be observed as it courses along in the body, it does not seem out of the ordinary that the corpuscles (which normally are quite flexible) change shape or contour as they crowd into interstices among the tissues or the cells thereof. Bowerbank, 1834, states that the spindle- or oat-shaped corpuscles become globular or spherical upon exposure to air.

Edwards, 1835-6, and Landois, 1864, believed that the blood corpuscle forms vary among different species of animals, but at all times appear to be essentially the same for postembryonic individuals of the same species. Prevost and Dumas state that the form and the volume of blood corpuscles are species specificities. Griffiths, 1892, stated that the sizes of blood corpuscles vary in the same individual.

Edwards, 1835-6, saw that the blood corpuscles of invertebrates are of very irregular form, surfaces rugose and readily change shape.

Now it is believed that but two classes of formed objects occur in the blood of insects; foreign bodies, such as parasites, bacteria, etc.; and blood corpuscles and their derivatives.

Usually there are but few kinds of corpuscles found in the blood of insects. Karawaiew, 1898, saw but large and small amebocytes, Bruntz, 1908, in Thysanura found four, Berlese working chiefly with muscids found four or five, Cuénot, 1895, working with Orthoptera reported four, Hollande, 1911, working with Coleoptera, Lepidoptera, Hemiptera, Hymenoptera, etc. found but three, Tillyard, 1917, working with Odonata found but two, and Muttkowski, 1924, working with several different kinds of insects distinguished but two kinds. Most of the foregoing investigators believe that these so-called different kinds of corpuscles are but successive phases in the development of a single corpuscle, but as Cuénot, 1896, stated, the life-history of an amebocyte is difficult if not impossible to trace.

In order to better differentiate among the blood corpuscles of cockroaches it was necessary to modify certain blood stains. Giemsa's, Hasting's and Wright's were tried. Each of the two former should be freshly prepared just before use, for they deteriorate upon standing. Wright's stain gave most satisfactory results when the blood smear was air dried and then treated with five drops of the undiluted stain for thirty seconds, being sure that the stain is spread over the smear. To this then was added as much distilled water as the slide would hold, permitting this to flood the slide for five minutes. Then the smear was thoroughly washed by moving it up and down and sidewise in a beaker of distilled water, or it was thoroughly drenched with a jet of distilled water from an ordinary pipette. With a piece of blotting paper the smear was dried and then was placed upon a radiator to become perfectly dry. Next the smear was mounted in neutral Canada balsam. This method gave results which were fully as

satisfactory as any, the process was the least troublesome or involved of the lot, and if kept away from direct light in tightly stoppered bottles, Wright's stain kept well for over a year.

In general appearance and behavior the blood corpuscles of the cockroach, Figs. 10 and 11, most correspond to the leucocytes of higher animals. Among them, in these investigations differentiations have been made with regard to size, proportional size relationships of the nuclei to those of entire cells, a comparison of staining affinities or intensities of the nuclei to those of the general cytoplasm, and the extent to which the nucleus and the cytoplasm was vacuolated.

According to the differentiation criteria as above set forth, it seems that there are several stages through which the corpuscles normally pass from the most recently divided to the farthest depleted condition, but these stages are not so easily separated as at first it may appear, for the phases gradually merge one into the other. A corpuscle in any stage may be more or less spent or vacuolated, but usually the vacuolations are more pronounced in the larger or older corpuscles.

Stage 1, Fig. 11, cytoplasm mediumly dense, rather heavily stained; nucleus rather heavily stained, rather large as compared to the cytoplasmic bulk, granules coarse, nucleus may or may not be in the center of the cytoplasmic mass. Usually the vacuoles of the cytoplasmic and nuclear masses were comparatively few and small. The most active and the most numerous of all corpuscles observed. Nuclear and general cell limits are as set forth in table No. 8. In a count of five hundred corpuscles made at random in an ordinary thin blood smear, three hundred seventeen were of this type. Fig. 1a of Fig. 11 shows one of these corpuscles in the division process; in the above count but two cells were seen thus engaged.

Stage No. 2, Fig. 11, seems to be a corpuscle of stage 1a after division. In it the nucleus is rather large as compared to the cytoplasmic mass, the cytoplasm showing as a narrow periphery around the nucleus. Usually the nucleus and the cytoplasm are rather densely stained, although the cytoplasm always is the fainter colored and sometimes may be but little apparent. The nucleus stains very heavily and its granules are large. These cells appeared to be rather active in the dark field. The nuclear and general cell limits are as given in table No. 8. In a count of five hundred corpuscles made at random in an ordinary thin blood smear, one hundred fifty were of this type.

Stage No. 3, Fig. 11, shows the appearance of corpuscles in the largest and last stage. The cytoplasm and the nucleus are highly vacuolar, stain but faintly, but usually the nucleus stained the darker. Of a cell in the far depleted or far spent condition, usually the nucleus appeared greatly reduced in comparison to the cytoplasmic mass, and the nucleus may or may not be central therein. The limits of size observed are as shown in table No. 8. In a count of five hundred corpuscles which was made at random in an ordinary thin blood smear, but eight were found which were of this type.

No. 4 of Fig. 11 shows what may be cockroach blood platelets. The nucleus and the cytoplasm are rather heavily stained, nuclei are regularly or irregularly outlined and may be fragmentary or entire. The sizes are as recorded in table No. 8. In a count of five hundred corpuscles which was made at random in an ordinary thin blood smear but twenty-nine of these forms were recognized. Their dimensions are as shown in table No. 8.

TABLE No. 8.

Cell and Nuclear Sizes.			Cell and Nuclear Sizes.		
Stage No. 1.	{	8 x28.8 9.6	Stage No. 2.	{	11.2x16 9.6x11.2
		12.8x16 8 x 9.6			12.8 9.6
		12.8x28.8 9.6x11.2			12.8x14.4 11.2
		14.4x20.8 9.6			12.8x16 8 x 9.6
		14.4x22.4 9.6x12.8			12.8x16 9.6
		14.4x25.6 9.6x12.8			12.8x19.2 9.6
		16 x17.6 11.2			12.8x19.2 9.6
		16 x19.2 9.6			16 x14
		16 x19.2 11.2x12.8			16 14
		16 x24 9.6			16 x17.6 9.6
		17.6x22.4 9.6			
		18.1x20.8 9.6x11.2			
Stage No. 1a.	{	18 x22.4 9.6	Stage No. 3.	{	22.4x32 9.6x12.9
		19.2 9.6x12.8			25.6x28.8 9.6
		19.2x20.8 9.6x12.8			25.6x32 9.6
		19.2x28.8 12.8x16			26.2x32 9.6
		20.8x22.4 9.6x12.8			32 9.6x12.8
		20.8 9.6x11.2			35.2 9.6x12.8
		22.4x28.8 9.5x15			38.4x41.6 9.6
		14.4 9.6			8 x13.4 9.6
		14.4 9.6			9.6x12.1 8
					9.9x12.8 obscure
					10.2x 8 8
		Stage No. 2.			{
9.6x14.4 6.4x 9.6	11.2 9				
9.6x20.8 9.6x12.8	11.2x12.8 obscure				
10.2x16 9.6x10.2	12.8 9.6				
11.2x16 9.6					

From what has been seen, it is believed that only the more vigorous corpuscles which have reached certain developmental phases divide to give rise to other corpuscles. Immediately or very soon after separating, it seems that the daughter corpuscles undergo a condition in which the nucleus becomes rather large in comparison to the general cytoplasmic mass, for in the resultant cells the nuclei are by far the most conspicuous parts, so filling the cell that the general cytoplasm thereof appears as but a narrow periphery about the nucleus. Apparently this stage is followed by one in which the general cytoplasm proportionally increases in volume, and thus the cell reaches its prime of growth, developmental and functional vigor. After this it may divide and the resultant daughter cells may grow to their prime of developmental and functional vigor, probably repeating the division process as long as they are sufficiently vigorous so to do.

Evidently the division process is a rapid, infrequent one, otherwise it would seem that corpuscles thus behaving would be more numerous in the mounts. Apparently the corpuscles live in their height of developmental and functional vigor for some time, for such by far are the most numerous in the blood.

It appears that gradually each corpuscle grows to maturity and that continuous functioning results in its becoming highly vacuolated, functionally debilitated or incapacitated and finally completely inactive. The writer believes that degeneration parallels the growing and functioning processes of every stage or phase of existence of the blood corpuscles and that up to a certain stage these degeneration processes are as gradual as are those of cell growth, but that when the degeneration processes have advanced to a certain stage, cell decline and wasting away to final disappearance must be greatly accelerated, and that probably on this account the corpuscles in later degeneration stages seldom are seen.

From what has been observed, the writer is convinced that the accompanying diagrams picture the successive phases or stages through which an individual corpuscle from cockroach blood ordinarily and normally passes during its existence.

With regard to blood corpuscle multiplication there are differences of opinion. Landois, 1864, says that they multiply by fission; Wheeler, 1892, states that in post-embryonic insects they numerically increase by mitosis; Cuénot, 1897; Henneguy, 1904; Bruntz, 1908; Hollande, 1911, and Tillyard, 1917, state that they divide both directly and indirectly. During my investigations I

have not seen mitosis, although I have rather closely examined several hundred smears.

Landois, 1864, regarded the pseudopods of blood corpuscles as albumin deposits. Crawley, 1909, saw thorn-like pseudopodia upon erythrocytes; Muttkowski, 1924, saw similar ones upon insect blood corpuscles. My investigations have revealed such structures upon the blood corpuscles of *Blattella germanica* Linn., and they are shown in Fig. 10, B and C.

Tillyard, 1917, states that he found but a few hundred blood corpuscles in a naiadal odonate. In the blood of the cockroach they are much more profuse. From an individual which had been starved for twenty-four hours there were about fourteen hundred to the medium-sized droplet. Fifteen of such droplets were available, and upon the foregoing basis, assuming that the droplets were of about the same size, in the fifteen there were about 21,000 corpuscles. Now in bleeding a cockroach probably not more than 50 per cent. of the total blood content can be directly bled out, the remainder being held in the interstices among the flaccid tissues which it bathes, so that by doubling the above number it is assumed that a conservative estimate of about 42,000 corpuscles is reached. A similar droplet was secured from a second individual. In it about 1,600 corpuscles were counted. Sixteen of such droplets were available, giving an approximation of 51,200 corpuscles content, calculated as previously stated. In a droplet from still another individual which had greedily eaten in the natural habitat, 2,480 corpuscles were counted in a single droplet. She yielded twenty of such droplets, which according to the previous calculation methods would give 99,200 corpuscles in her blood. From the foregoing it is seen that there are wide variations in blood corpuscle content in individuals of the same species, and seemingly those which are well nourished and recently have fed have more blood and a greater number of corpuscles in it per unit volume. It is safe to state that at least thousands of corpuscles normally occur in the blood of a cockroach.

From what has been seen the investigator believes that all cockroach blood constituents contribute to coagulum formation, for when favorably exposed for that purpose, the irritated or stimulated corpuscles send out pseudopodia as is shown in A and B of Fig. 10. It long has been known that insect blood clotting in large measure consists of the interlacing and more or less complete fusing of contiguous portions of the pseudopodia which the cor-

puscles send out. The fibrinogen acts to generate fibrin which clogs the interstices or meshes occurring among the pseudopodia, so that fibrin is a coagulum constituent. Later, seemingly the corpuscles, their pseudopodia and the fibrin contract, and as a result the liquid serum is squeezed from the clot. If the fresh coagulum is completely lifted from the slide, it will be seen that the surface formerly covered by it still is moist. This moisture is due to serum which was squeezed from the contracting clot as it further dried. To the liquid serum the moisture of the coagulum is due, and it is chiefly the liquid serum which evaporates when the coagulum shrinks and dries to hardness. The watery fluid or liquid serum is blood plasma which is devoid of corpuscles, fibrinogen and fibrin, and to it the fluidity of normally circulating blood is due. Ordinarily liquid serum covers the surface of the fresh clot. If ordinarily exposed, after a time this serum dries down to fuse or mat the clot surface. As the latter further dries it hardens, taking on a smooth glazed or glossy appearance. In the thoroughly dried and hardened coagulum the corpuscles have lost their identity and numerous crystals appear throughout the dried mass.

In Ringer's solution of ordinary composition, cardiac action continued for six hours and about eighteen minutes; in a .6 gram sodium chloride solution in which other salts were reduced accordingly, the heart continued functioning for two hours and about thirty minutes. When the solution ordinarily was made up, with the exception of omitting the dextrose, in one case heart action in it was sustained for five hours and about thirty-five minutes; in another case for four hours and about forty minutes.

Doubtless blood dust as extremely minute albumin and fibrin particles was seen in blood by Milne-Edwards, 1835-6; Stokes and Wegfarth, 1897, and Crawley, 1909. Stokes and Wegfarth, 1897, believed that it is of leucocytic origin, Nicholls, 1906, regarded it as of diverse origin and Crawley, 1909, considered it of erythrocyte origin. The investigator has seen it in the blood of cockroaches. There an erythrocyte origin for it is out of the question, and the writer believes that it is derived from the leucocytes in insect blood.

LIPOMICRONS

Gage and Fish, 1921, experimenting with higher animals found that the fat of the diet appears in the blood as a reconverted material, a fat which is reconstructed from its constituents before it has had an opportunity to enter the blood as species fat. With

the dark field microscope they saw freely floating micro fat particles which were carried by the blood stream after fat had been included in the diet. They mention that differences in the form of fat curves (constructed from data secured on digestive activity from hour to hour during a twenty-four hour interval) may be expected in different species as well as in the same individual at different times. In horse, dog, cow, etc., they saw that there are waves of fat absorption during the twenty-four hours following the feeding of fat. They showed that if only certain constituents of fat (fatty acids or glycerols) are fed, certain cells of the digestive tract wall supply the remaining constituents which are necessary for fat synthesis, and that as a result micro fat particles (lipomicrons) are liberated into the blood. Drs. Gage and Fish have traced such fat to fat reserves in the body, where it was stored for future use. Bloor, 1916, saw that lipomicrons occur in the blood stream of well nourished starving dogs. Green, 1914, working with the king salmon found that reserve fat is stored in various tissues of the body, and from such fat reserve stations was drawn upon as an energy source for the spawning migration, during which no food is taken. It has been demonstrated that lipomicrons appear in the blood of the German or Croton cockroach, *Blattella germanica* Linn., after fat or either of its constituents has been included in the diet. The curves of Fig. 13 further deal with the appearance of fat in the blood of these insects which were especially fed for the purpose.

When the average temperature and relative humidity were 20.4 degrees C. or 68.7 degrees F. and 87.9 per cent. respectively over a twenty-four hour interval, the height (maximum) of lipomicron appearance was reached in from 19 to 19½ hours; when the average temperature and relative humidity were 23.6 degrees C. or 76.4 degrees F. and 85.5 per cent respectively during a twenty-four hour interval, the maximum of lipomicron appearance in the blood was reached in from 14 to 19 hours. After twenty-two hours and up to forty hours at an average temperature of 22.5 degrees C. or 72.5 degrees F. and a relative humidity of 91.2 per cent., fat digestion was about the same as during the interval before the maximum was reached.

Curves 19-21 inclusive of Fig. 13 are recorded from starving individuals as compared to Nos. 16-18 inclusive, in so far as fat is concerned. Apparently the individuals of the latter group were drawing from the lipomicrons directly from the digestive tract, while those of the former were drawing from the fat reserve storage stations of the body.

TRACHEAE AND SPIRACLES OF COCKROACH.

Nothing especially striking has been revealed in dissections to demonstrate the tracheal system. It may be well to mention that freshly killed specimens were impaled upon a very fine pointed pipette which was made by drawing a piece of small soft glass tubing to a point. The fine point was thrust through the body wall between the first and second pair of legs. Specimens then were inflated by blowing into the larger end of the pipette until the normally concealed genitalia bulged forth, an excellent scheme for revealing such structures in freshly killed specimens. The inflated individual, still impaled upon the fine pipette and under air pressure from the mouth of the operator was held over an electric hotplate until thoroughly dried, known by occasionally releasing the pressure. At such times, if specimens slightly collapsed continued inflation and desiccation were necessary until upon releasing the pressure, the specimens remained thoroughly rigid. If thoroughly inflated and dried to the extent as above indicated, the intersegmental membranes are so stretched that any structures which they bear are bulged forth to be clearly revealed if properly examined.

There are ten pairs of spiracles, but unless specimens are especially prepared, all but the last abdominals are difficult to locate. The tracheal system consists of large, parallel tracheal trunks which are connected by cross commissures. Dilatations may occur in the main trunks, in the commissures or directly or indirectly connected with either or both. By some, these dilatations have been mistaken for air sacs, but taenidia are developed in them. It is doubtful that any two tracheal systems are exactly alike; in fact rarely are the two sides of the tracheal system of the same individual exactly symmetrical. Occasionally rather well developed portions in one individual correspond to rather poorly developed portions in others, or parts which are present in one individual may not be represented in another (Figs. 14-20).

In some insects air may be taken into other than the tracheal system, as has been observed in the cockroach. Just previous to the molt specimens have been noticed to bob the front end of the body up and down. In the meantime the body, especially the abdomen, becomes so distended that it loses its flat appearance and becomes more cylindrical. Soon the muscles upon each side of the thoracic terga begin to contract and relax, and at the same time a definite line or crease appears along the middle of the

length of the dorsum of each thoracic segment. This continued until the old cuticula finally separated along that line similarly as does a piece of tin which is repeatedly creased and uncreased by folding and unfolding along the same line. The specimen then molted. After the wings had begun to unfold, their tips were snipped and several drops of blood came from them, but comparatively little came from snipped antennal and leg tips. An incision then was made through the dorsal surface near the posterior end of the abdomen. Through this incision an inflated membrane appeared. The projecting portion was stained with alcoholic eosin, and upon dissection it was revealed that it was the crop which was air inflated to the extent that it crowded the remainder of the alimentary tract into the posterior abdominal extreme. Thus via the crop air pressure aided in rupturing the old cuticula and exerted pressure upon the blood which aided in unfolding the elytra and the true wings. In other individuals blood could not be obtained from the wings after the body normally had become flattened and of color characteristic of the adult. If the crop is punctured while the body is in the inflated cylindrical condition, the ruptured structure collapses as does an inflated tire at the time that it is punctured. Knab, 1909-1911, noted a similar function of air in flies and other insects.

SUMMARY AND CONCLUSIONS.

The average percentage of moisture in an adult cockroach from the natural habitat is about 68, and assuming that about one-third of it may be blood, gives about 20 per cent. of the body weight due to its blood content. But this may greatly vary due to the nutrition status of the individual.

The proportion of moisture to body weight is still higher in younger individuals. Doubtless a great portion of the moisture is present in the form of blood plasma and tissue serum, the latter a derivative of the former and practically inseparable from it, so that the amount of blood as such which occurs in the insect body never has been satisfactorily calculated. Temperature and relative humidity influences as well as the nutrition status have their effects upon the moisture content of the body, for well nourished individuals contained more blood than did those which were completely starved for twenty-four hours or longer; and cockroaches which had lost much metabolic water as a result of having been exposed to unfavorable relative humidity and thermal influences yielded blood with as great difficulty as did those which had

been completely starved for some time, so that the metabolic water must have been yielded from the blood and probably from the tissues. If the individual is deprived of solid food but has access to sufficient water, it bleeds readily and profusely until it has starved to death.

At lower temperatures cockroaches of this species seek seclusion in places the relative humidity of which is higher; at lower relative humidities they sought seclusion in places the temperatures of which were higher.

Of all Orthoptera, Homoptera, boring larval Lepidoptera, two phytophagous larval Lepidoptera and two Coleoptera, one of the family Cerambycidae, the other of the family Carabidae, the blood reacted alkaline to neutral litmus.

Of all phytophagous Coleoptera, pollen eating Coleoptera, phytophagous and pollen eating Coleoptera, carnivorous Coleoptera and phytophagous larval Lepidoptera but two species, the blood reacted alkaline to neutral litmus.

Of all Diptera which were examined, the blood reacted alkaline to neutral litmus.

The blood of certain phytophagous larval Lepidoptera reacted alkaline, while that of certain others reacted acid to neutral litmus.

The blood of a pollen eating beetle, *Cyllene robiniae* (Cerambycidae) reacted alkaline, while that of others, *Chauliognathus* (Lampyridae) and of *Trirhabda* (Chrysomelidae) reacted acid to neutral litmus.

The blood color of representatives of different families of the same order of insects may differ, although the individuals feed upon the same host species. Among insects blood color never is due to that of the corpuscles, but always is due to that of the plasma.

The blood of certain carnivorous Coleoptera (Coccinellidae) reacts acid, while that of others (Carabidae) reacts alkaline. The blood of the former is of reddish color, while that of the latter is of turbid whitish color.

The blood of adult phytophagous Coleoptera which were examined is of yellow color, while that of phytophagous larval Lepidoptera usually is of greenish color.

Blood color modifications are of little taxonomic importance.

The results of these experiments reveal that influences other than host plant sap act in the coloring of insect blood, for insects of different species feeding upon hosts of the same species have blood which may present color differences and which may be of

different color than is that of the host sap or tissues upon which the insects feed; that insects of different species feeding upon hosts of the same species may have blood which reacts differently and that the reaction of blood to neutral litmus usually is different than that of the sap or tissues which serve as food.

Ordinarily larvae bear more blood in proportion to the body weight than do adults.

From the foregoing investigations it is further concluded that the acidity, alkalinity and color of insect blood are species specificities and are influenced by more than just the food which the insect takes.

Ordinarily the constituents of the plasma and those of the corpuscles are difficult to separate by hard and fixed lines, for it seems that some of the constituents of each are interchangeable.

The corpuscles of cockroach blood are not known to be of respiratory character, although very locally the plasma may be concerned with the transportation of respiratory gases.

It is believed that the blood of the cockroach is concerned with the transportation of nourishment as the chief function. This it carries to and among the tissues, removing metabolic wastes from them. It is doubtful that the blood corpuscles carry food to the tissues, at least microparticles of fat profusely and freely occurred in the blood stream.

All individuals did not bleed equally readily, some bled more easily and more profusely from clipped antennae than they did from clipped cerci and vice versa. Some at times bled more profusely and more easily than they did at others.

The basic colors of fresh and of dried cockroach blood are very similar.

According to spectroscopic and biochemic analyses the blood of cockroaches seems to be as complex as is that of other animals.

There are several kinds of crystals in the blood of cockroaches, and it appears that they are of organic nature.

In coagulation it seems that all blood constituents of the cockroach take part, for in the process the corpuscles send out pseudopodia which fuse with those of other corpuscles, and fibrin clogs the interstices among them. The mass later contracts and as a result the serum is squeezed out.

Reflex bleeding is not recorded among cockroaches. Cockroach blood is neither toxic nor repellant to certain kinds of spiders, assassin bugs and other arthropods.

Ringer's solution of .8 per cent. sodium chloride concentration gave best results in sustaining cardiac action, although good re-

sults were obtained when a .6 per cent. concentration of sodium chloride solution with reduction of other salts accordingly was used, whether dextrose was or was not included.

Lipomicrons appear in the blood of especially fed cockroaches similarly as do chylomicrons in the blood of higher animals.

Blood dust similar in appearance and behavior to that in higher animals is found in cockroach blood. In insects it is highly probable that it comes from the blood corpuscles.

Peculiarities in the behavior of cockroach blood corpuscles paralleling those which have been seen in the blood of higher animals have been observed.

Wright's stain gave the most satisfactory results of those which were used in working with cockroach blood corpuscles. The process was the least troublesome or involved of the lot, the results were more permanent and the stain kept well for over a year if placed into tightly stoppered bottles which were stored away from direct light.

In behavior, appearance, etc. cockroach blood corpuscles are very similar to the leucocytes of higher animals. It is believed that only the more vigorous corpuscles which have reached certain development phases can divide to give rise to other corpuscles. Immediately or very soon after separating it seems that the daughter corpuscles undergo a condition in which the nucleus is rather large in comparison to the general cytoplasmic mass, for in the resultant cells the nuclei are by far the most conspicuous parts, so filling the cells that the cytoplasm thereof appears as but a narrow periphery about the nucleus. Apparently this stage is followed by one in which the general cell cytoplasm proportionally increases in volume, and that thus the cell reaches its prime of growth, developmental and functional vigor. After this it may divide and the resultant daughter cells may grow to their prime of developmental and functional vigor, probably repeating the division process as long as they are sufficiently vigorous so to do.

Evidently the division process is a rapid, infrequent one. Apparently the corpuscles live in their height of developmental and functional vigor for some time, for such by far are the most numerous in the blood.

It appears that gradually each corpuscle grows to maturity, and that continuous functioning results in its becoming highly vacuolated, functionally debilitated or incapacitated and finally completely inactive, soon after which it entirely wastes away or disappears. The writer believes that degeneration parallels the grow-

ing and functioning processes of every stage in the existence of the blood corpuscles and that up to a certain stage these degeneration processes are as gradual as are those of cell growth, but that when the degeneration processes once reach a certain condition, the cell decline and wasting away to final disappearance must be greatly accelerated, and that probably on this account corpuscles in the later degeneration stages seldom are seen.

From what has been observed, the writer is convinced that the accompanying diagrams, Fig. 9, picture the successive stages through which an individual corpuscle from cockroach blood ordinarily and normally passes during its existence.

There are at least thousands of blood corpuscles in an adult cockroach. It is believed that the total volume of corpuscles as compared to that of the blood plasma varies in the same individual from time to time and that one individual may contain more blood than does another. It is believed that the nutrition status of the individuals influences the amount of blood which that individual contains.

If the temperature is too low in the cockroach, fat digestion may be arrested, although fat is present in the crop.

In well-nourished, starving cockroaches it seems that fat is withdrawn from fat reserve stations of the body.

In my experiments fat has been demonstrated only in the free state (as lipomicros) in the blood stream.

Somewhat as in higher animals, waves of fat digestion occur in the Croton or German cockroach, *Blattella germanica* Linn.

Differences in the forms of curves may be expected in individuals of the same species, although they are subjected to identical thermal and relative humidity influences.

Ordinarily in the cockroach the time of maximum appearance of fat in the blood in a rough or general way approximates that of certain higher animals.

If fatty acids or glycerol alone are fed, the tissues of the digestive tract wall supply the necessary constituents (fatty acids or glycerols) to synthesize species fat. It appears that fat digestion in this insect is as much of an involved process as is that in a higher animal.

These researches tend to indicate that the blood of insects is as highly complex a tissue as is that of the higher animals.

Nothing particularly striking has been revealed in dissections which were made to demonstrate the tracheal system in its grosser aspects. There are ten pairs of spiracles. Certain tra-

cheal dilatations have been mistaken for air sacs, but taenidia are developed in them. Usually the two sides of the tracheal system of the same individual are not exactly symmetrical. Occasionally rather well developed portions in one individual correspond to rather poorly developed parts in others, or parts which are present in one individual may not be represented in another. Air pressure brought about by a fully distended or inflated crop aids in making the molt in cockroaches, and it is believed that this exerts a pressure upon the blood in unfolding the elytra and true wings at the time of the final molt.

From the main tracheal trunks smaller branches are given off, these still more finely divide, and so on down to the finest or most minute. These latter are reduced to microscopic capillarity (less than a micron in diameter). So a portion of a tracheal trunk showing all attached derivative branchings to the most minute extremes presents a dendritic arrangement, the minutest extremities of which are so fine and so numerous that but few if any of the cells of the tissues which are aerated by the portion under consideration are not reached or permeated by them.

CAPTION TO FIGURES.

Figs. 1-3. Cockroach trap, showing method of making the trapping cone and placing it within a trapping receptacle, Fig. 1. In such a trap decoys of the species to be trapped are used; so are attractive baits. This type of trap catches cockroaches without injury; only uninjured individuals were used in these investigations.

Fig. 4. Type of bottle cage in which uninjured experimental material was kept during the investigations. The cork is perforated and the lower end of the perforation is screened with a fine meshed brass screen. A bit of dampened sponge is kept in the perforation so that the air in the cage is kept moist, as indicated by condensation upon the walls of the cage. A piece of pasteboard was cut sufficiently wide so that it just easily passed into the mouth of the bottle. Its length was slightly greater than the inside diameter of the cage. Upon introducing it into the cage, it was placed with the free ends directly downward and as it straightened out until the ends reached the walls it formed a shelter beneath which the specimens spent much of their time. Food was placed into the bottle as frequently as the specimens needed it.

Fig. 5. Temperature and relative humidity control tube the use of which has been explained in the text.

Fig. 6. Individual housing vial wherein the temperature and relative humidity were kept constant during experimentation.

- Fig. 7. Individual housing vials arranged in a constant temperature oven the heat of which was supplied by a 60 watt bulb, the humidity was kept constant by a vat of water, W; the bit of water saturated sponge in the perforated cork of the individual housing vial and by the water in the vial, shown by 5 beneath the glass plate 4 of the vial. .
- Fig. 8. Demonstrates my method of making blood smears so that the blood corpuscles are little if at all injured in the process.
- Fig. 9. Demonstrates different types of blood crystals of cockroach blood.
- Fig. 10. Cockroach blood corpuscles in action in a fresh blood smear.
- Fig. 11. Different types of cockroach blood corpuscles.
- Fig. 12. Photograph of the spectrum which was produced as dried cockroach blood was burned in investigating the mineral contents thereof.
- Fig. 13. Shows the curves which were obtained by recording the numbers of lipomicrons which were present at various time intervals after the experimental individuals had been fed fat. The numbers at the ends of the curves indicate the individual concerned in obtaining the data; the figures on the horizontals are time interval notations, recorded by the hour and by the half hour.
- Fig. 14. Spiracular distribution upon the cockroach body.
- Fig. 15. Dorsal position of the first pair of abdominal spiracles.
- Fig. 17 (and that above). Lateral views of the tracheal system of a cockroach, *Blattella germanica* Linn.
- Fig. 18. One of a dorsal tracheal arrangement of an individual *Blattella germanica* Linn.
- Fig. 19. One of a ventral tracheal arrangement of an individual *Blattella germanica* Linn.
- Fig. 20. Scheme of tracheal arrangement in a cross section taken through the spiracle region of one of the abdominal segments.

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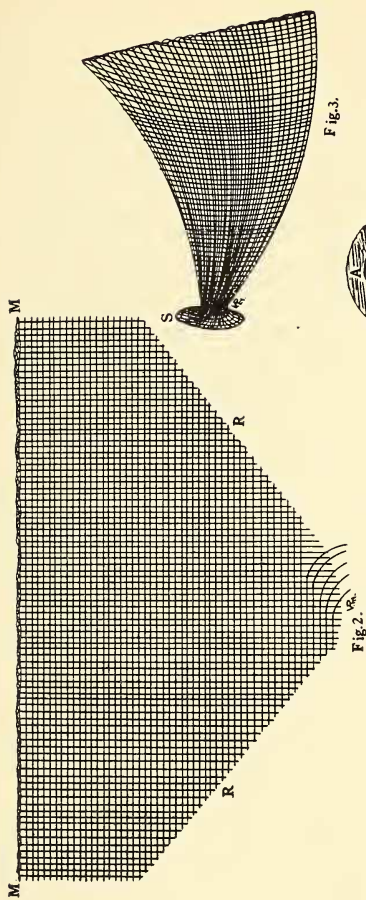


Fig. 3.

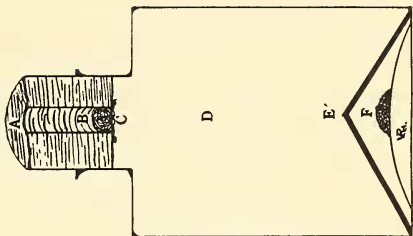


Fig. 4.

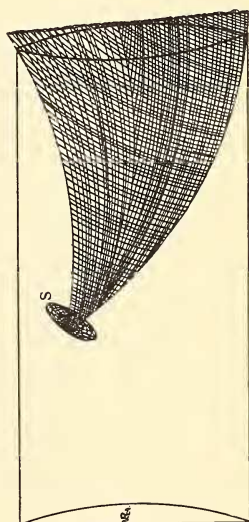
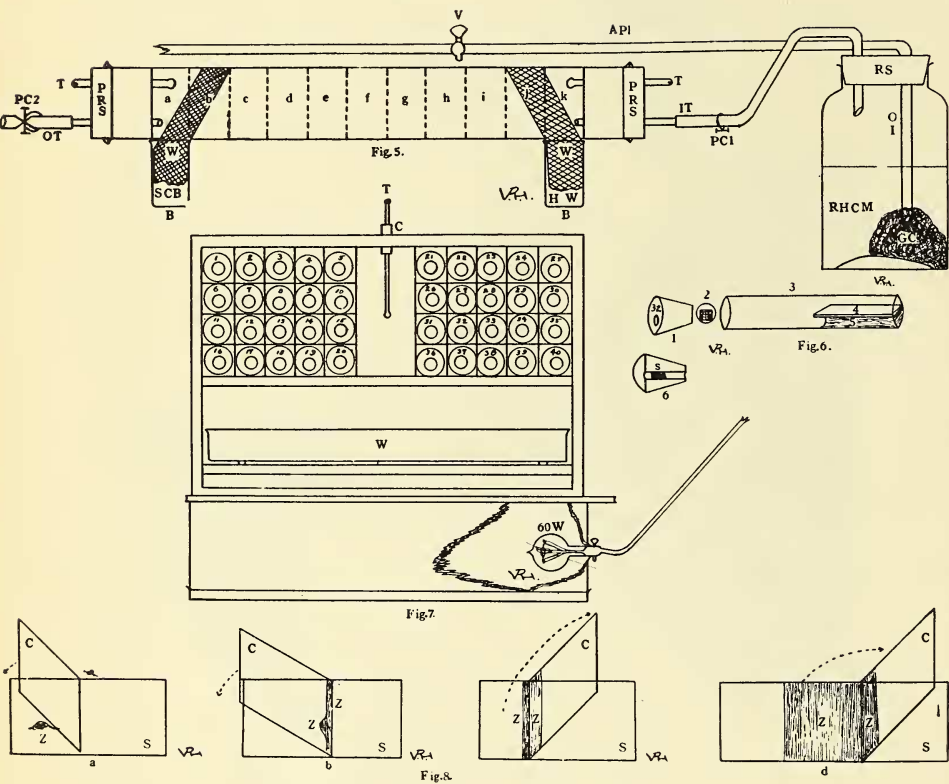


Fig. 1.



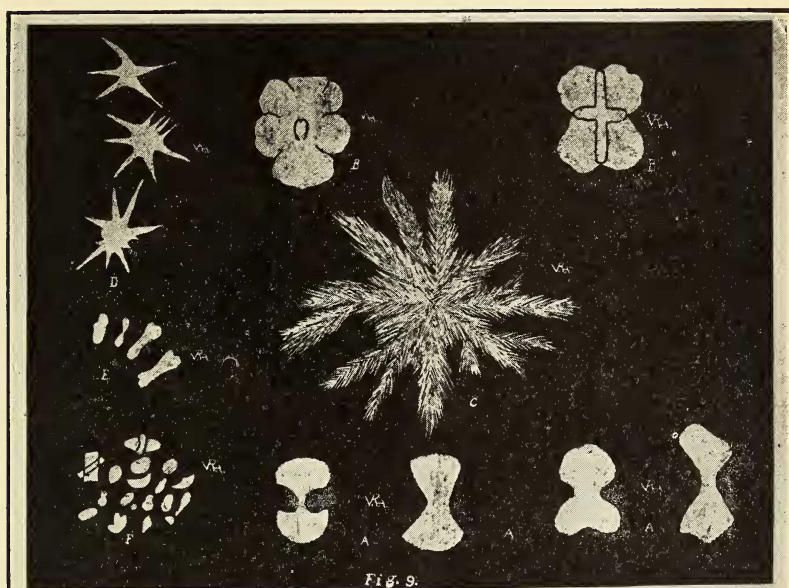


Fig. 9.



Fig. 10.

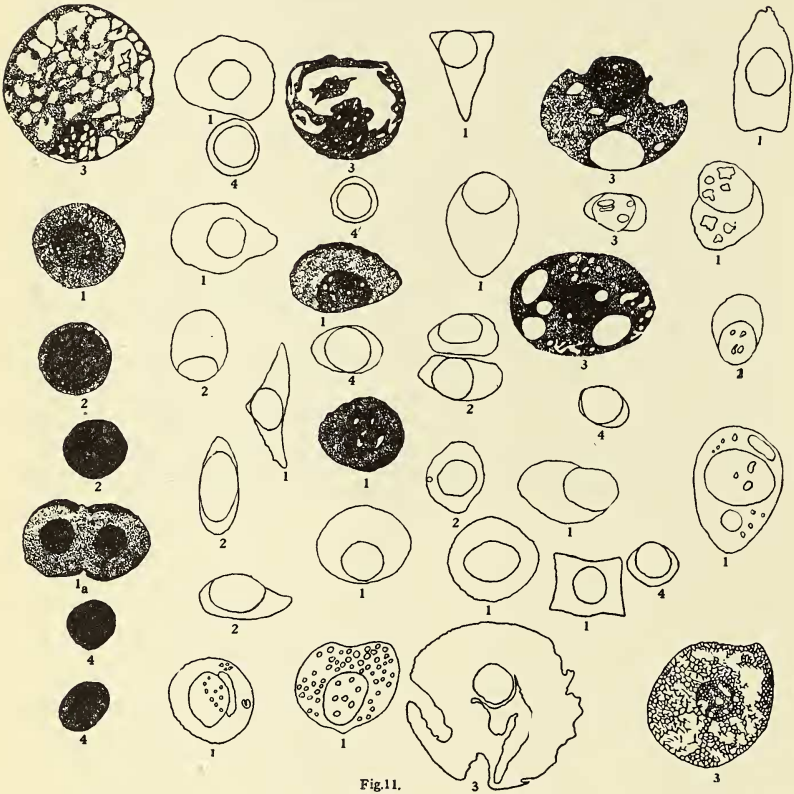
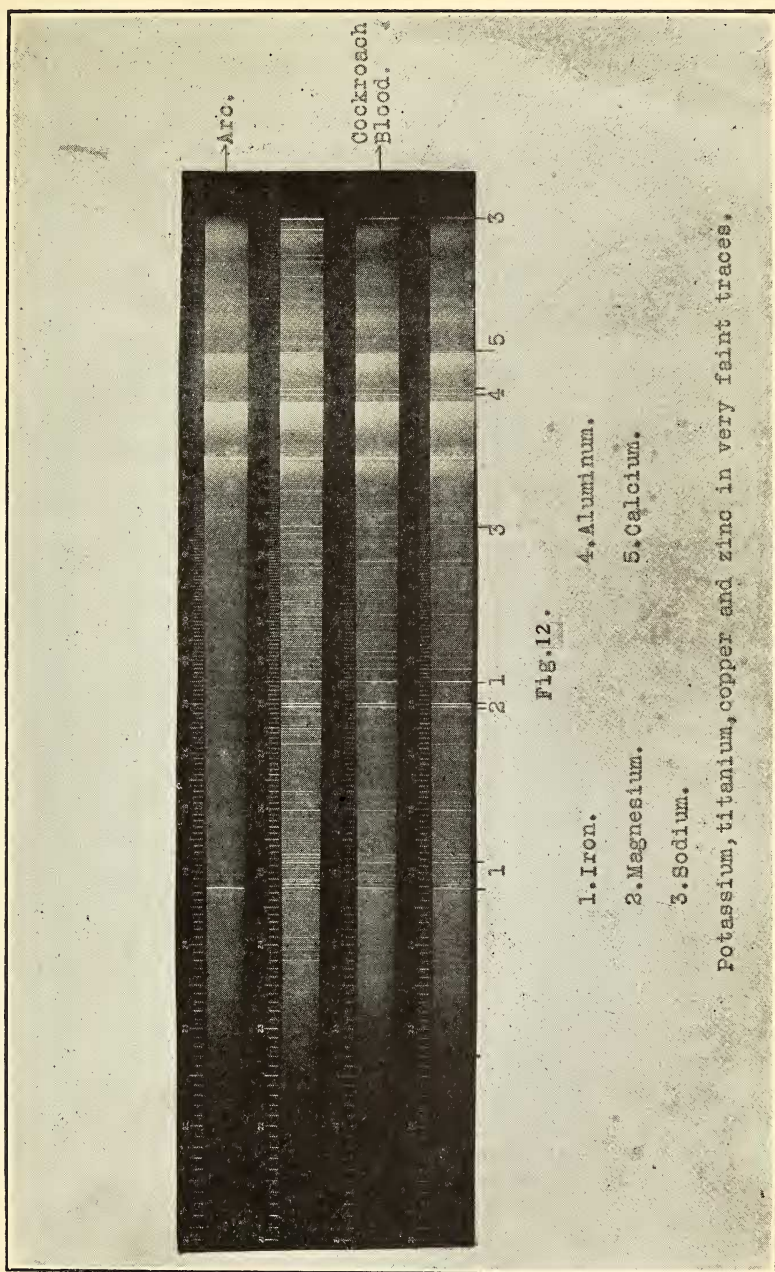
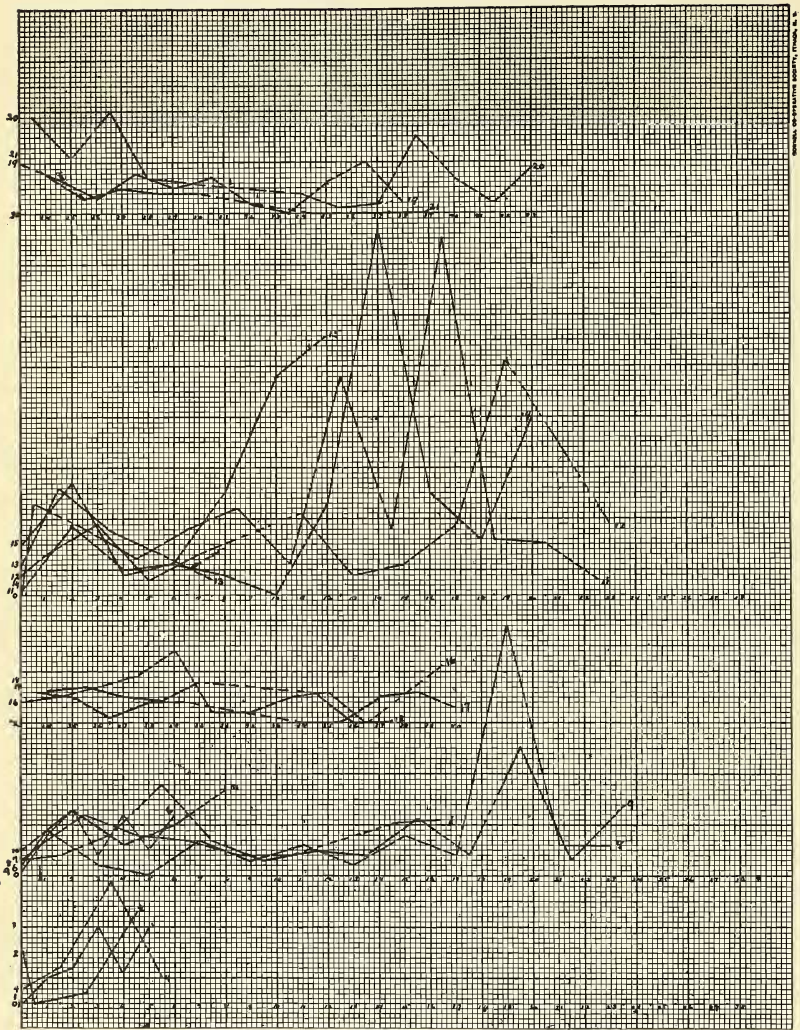


Fig.11.





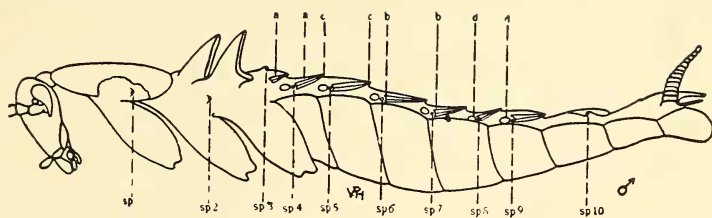


Fig. 14.

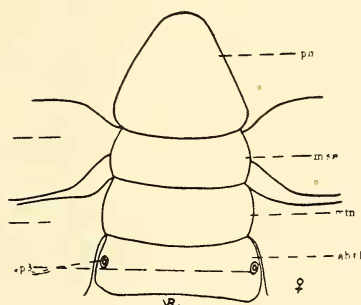


Fig. 15.

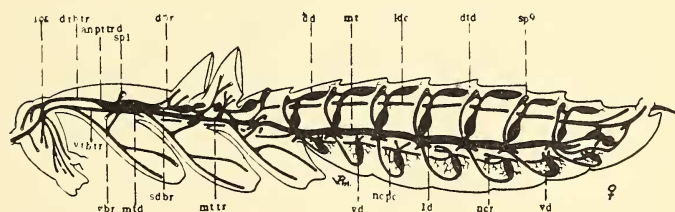
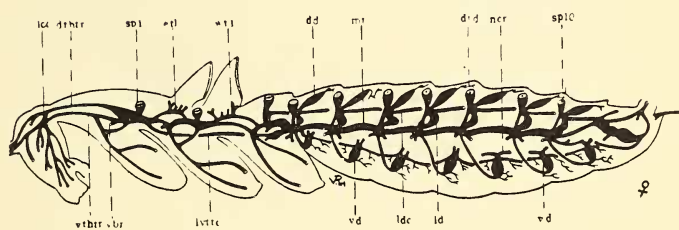


Fig. 17.

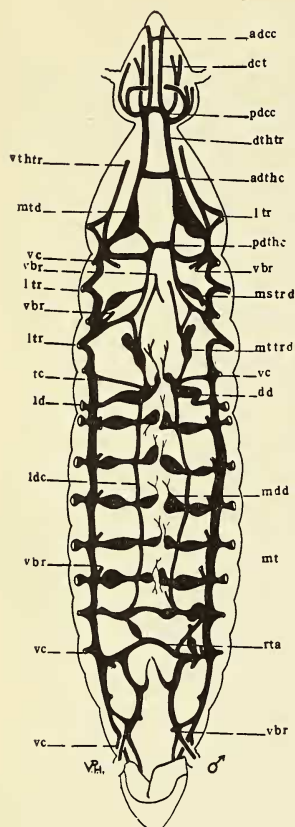


Fig. 18.

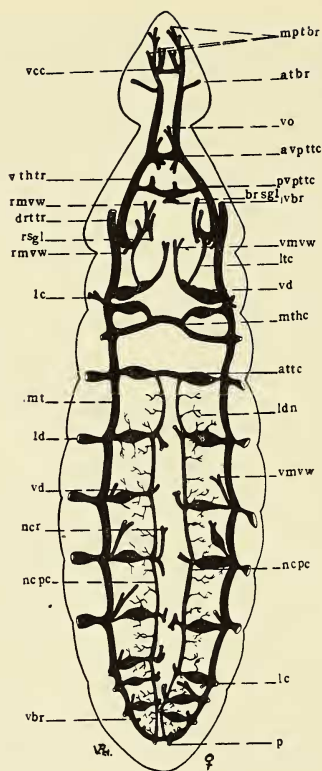


Fig. 19.

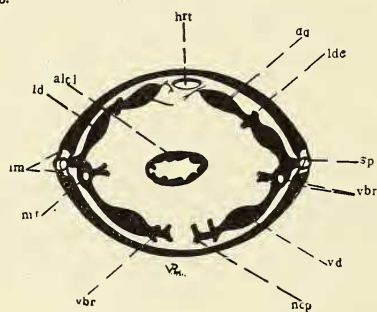


Fig. 20